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# A HISTOLOGICAL STUDY OF THE DUAL AFFERENT INNERVATION OF THE ESOPHAGUS OF THE DOG

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by

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## INTRODUCTION

LANGLEY sectioned the dorsal root distal to its ganglion and found that all the myelinated fibers in the corresponding white rami communicantes had degenerated. Section of the sympathetic trunk or the splanchnic nerve did not cause such degeneration. On the basis of these observations, LANGLEY concluded that the visceral afferent fibers pass through the sympathetic trunk.

RANSON and BILLINGSLEY presented evidence which indicates that the visceral afferent fibers of the sympathetic trunk are chiefly myelinated fibers of large and medium size, but small myelinated and unmyelinated visceral afferent fibers also occur.

EDGEWORTH, FOERSTER, FULTON and KUNTZ have recognized the existence of the visceral afferent fibers in the sympathetic trunk from the histological standpoint. SHEEHAN found that the nerve fibers in the VATER-PACINIAN corpuscle in the mesentery have cell bodies in the dorsal root ganglia. BALCHUM, WEAVER, EBEDENKO, BRJUSSOWA, CANNON, BAIN, IRVING, MCSWINEY, SUFFOLK. ISHIKAWA, KUBO, ASAI, HIRAMATSU and NOSE have maintained from the physiological standpoint that the visceral afferent fibers pass through the sympathetic trunk or the dorsal root.

NEUMAN, BROWN, IRVING, MCSWINEY, SUFFOLK, FULTON, WHITE, SMITHWICK, MELTZER, AUER, KUBO, ASAI, MIZUTA and NOSE presented physiological evidence that the vagus contains visceral afferent fibers. RANSON, FOLEY, and ALPERT have maintained the evidence of the vagal afferent fibers from the histological standpoint.

SADA found afferent nerve endings in the esophagus, but he did not try to find the source.

Therefore, it is evident that the vagus and the sympathetic nerve play important rôles in conducting the afferent impulses from the esophagus. But I have not found literature on histological studies of this subject, so I attempted to solve the problem histologically.

## MATERIAL AND METHODS

Adult dogs were used. Preparations of the esophagus of a normal dog were stained with BIELSCHOWSKY-SETO'S silver method and EHRLICH'S acid hematoxylin method. And the secondary degeneration of the nerves in their esophageal tissues were investigated by BIELSCHOWSKY-SETO'S silver method and MARCHI'S chromosmium method.

I experimented as follows :

- (1) Unilateral cervical vagotomy at a point distal to ganglion nodosum.
- (2) Section of the dorsal roots of both sides at points proximal to their ganglia (Th. 4-Th. 8).
- (3) Section of the dorsal roots of both sides at points distal to their ganglia (Th. 3-Th. 7).
- (4) Unilateral phrenic exeresis.

The dogs, in whom cervical vagotomy had been performed on both sides at the same time, died of dyspnea, etc.. Therefore, vagotomy was performed on one side, but I did not find a marked difference between the microscopic observations of nerves of the esophagus after either the right or left vagotomy.

First, degeneration of nerve fibers was investigated by BIELSCHOWSKY-SETO'S method in each dog, which was killed 25, 12, 8, or 6 days after vagotomy. Degenerated nerve fibers were not found in the esophagus of any of the dogs killed 25 or 12 days after vagotomy. But I found a few degenerated fibers in each case where the dogs were killed 6, 7 or 8 days after vagotomy, especially in the case of dogs killed 6 days after vagotomy.

MARCHI'S stain was used to pursue the peripheral nervous pathways in the esophagus of each dog which was killed 21, 12, 9, 8, 7 or 6 days after vagotomy. MARCHI'S granules were found in each case where the dog was killed 9, 8, 7 or 6 days after vagotomy, and most markedly in the case of a dog killed 6 days after vagotomy. They were not found in the case of dogs killed 21 or 12 days after vagotomy.

Therefore, I searched for degenerated fibers in the esophageal tissue of each dog which was killed 6 days after nerve section.

#### MICROSCOPIC OBSERVATIONS.

Afferent fibers in the normal esophageal tissue of a dog.

Fig. 1 shows a tangled ending in the connective tissue between the muscle bundles. A nerve ending in Fig. 2 shows a network in an AUERBACH'S plexus. SADA and SETO have maintained that they are afferent endings.

I found myelinated fibers in the lamina propria of the mucous membrane by EHRLICH'S method (Fig. 3).

Degeneration of nerves in the upper part of the lower third of the thoracic esophagus, the preparation of which was stained with BIELSCHOWSKY-SETO'S method.

(1) Unilateral cervical vagotomy at a point distal to ganglion nodosum. Fig. 4 and Fig. 5 show fragments and vacuoles of degenerated nerve fibers in an AUERBACH'S plexus and in the muscle.

(2) Section of the dorsal roots of both sides at points proximal to their ganglia (Th. 4-Th. 8).

Degenerated nerve fibers are not found.

(3) Section of the dorsal roots of both sides at points distal to their ganglia (Th. 3-Th. 7).

Few degenerated nerve fibers are seen in the submucous tissue (Fig. 6).

Degeneration of nerves in preparations stained with Marchi's method.

(1) Unilateral cervical vagotomy at a point distal to ganglion nodosum.

MARCHI'S granules are abundantly found in an AUERBACH'S plexus in the upper part of the lower third of the thoracic esophagus (Fig. 7). There are also quite a few in the muscle layer (Fig. 8), in the submucous tissue (Fig. 9), and in the submucous plexus.

(2) Section of the dorsal roots of both sides at points proximal to their ganglia (Th. 4-Th. 8).

MARCHI'S granules are not found in the upper part of the lower third of the thoracic esophagus or in the abdominal esophagus.

(3) Section of the dorsal roots of both sides at points distal to their ganglia (Th. 3-Th. 7).

MARCHI'S granules are seen in the muscle layer (Fig. 10), in an AUERBACH'S plexus (Fig. 11), and in the nerve bundle which runs from this plexus into the submucous tissue (Fig. 12). These granules are found in small numbers in the upper part of the middle third of the thoracic esophagus, and in the abdominal esophagus, but they increase in numbers in the upper part of the lower third of the thoracic esophagus.

Where the dorsal roots have been sectioned at points distal to their ganglia, there are only about one tenth as many MARCHI'S granules in the upper part of the lower third of the thoracic esophagus as in the case of vagotomy.

(4) Unilateral phrenic exeresis.

MARCHI'S granules are not found in the portion of the esophaguns at the esophageal hiatus of the diaphragm.

#### DISCUSSION

In the neck of the dog, the vagus unites with the sympathetic trunk and it is difficult to separate them. Therefore, when the vagus is cut in this point, the sympathetic trunk is also sectioned. According to RANSON'S and BILLINGSLEY'S study of the cat, the cervical sympathetic trunk consists of few unmyelinated and many myelinated fibers. The unmyelinated fibers are postganglionic, rise in the superior cervical ganglion, and extend in the internodal ramus for a short distance before entering a peripherally distributed ramus. The myelinated fibers degenerate toward the superior cervical ganglion following section of the sympathetic trunk at any level in the neck, and the degenerating process does not extend downwards. Therefore, degenerated nerve fibers, if any, in the esophageal tissue following section of the cervical vago-sympathetic trunk must be considered vagal.

It has been thought that the cell bodies of the afferent fibers of the vagus lie in the ganglion nodosum (RANSON, LARSELL, CLARK). Therefore, degenerated fibers in the esophageal tissue following vagotomy are not always afferent, because many efferent myelinated fibers which have no synaptic connection on their way, supply the striated musculature of the esophagus (RANSON, LARSELL, KURE and OKINAKA). Myelinated fibers in the submucous tissue, however, do not supply the striated

musculature, because the muscularis mucosae is the smooth muscle. It can be supposed that efferent myelinated fibers enter the esophageal gland. But I could not learn from the references whether or not efferent myelinated fibers supply the esophageal gland. According to SMITH and others, the parasympathetic preganglionic fibers of the vagus end in the ganglia of the plexus of the alimentary canal. Therefore, even supposing that the efferent fibers of the vagus contain myelinated fibers in the submucous tissue, they must not undergo secondary degeneration following cervical vagotomy. The major portion of the submucous plexus in the esophagus lies close to the internal muscle layer, and many investigators have not found ganglia in the esophageal submucous plexus (KUNTZ). My findings were identical with theirs. Therefore, it seems reasonable to assume that the degenerated myelinated fibers which run in the submucous tissue at a distance from the internal muscle layer are afferent (Fig. 9).

It is not possible to distinguish whether degenerated nerve fibers in the muscle layer and AUERBACH'S plexus following vagotomy, as shown in Fig. 4, 5, 7 and 8, are afferent or efferent.

MARCHI'S granules, as shown in Fig. 10 and Fig. 11, and especially the large granules which reach into the submucous tissue, shown in Fig. 12, are found in the cases following section of the dorsal roots at points distal to their ganglia from Th. 3 to Th. 7. But they are not found in the cases following section of the dorsal roots at points proximal to their ganglia from Th. 4 to Th. 8. These facts indicate the existence of afferent fibers in the esophagus which have their cell bodies in the dorsal root ganglia.

Following section of the dorsal roots at points distal to their ganglia (Th. 3-Th. 7), MARCHI'S granules are few in the upper part of the middle third of the thoracic esophagus and in the abdominal esophagus, but they increase in numbers in the upper part of the lower third of the thoracic esophagus. The upper part of the middle third of the thoracic esophagus is about at the level of the dorsal root ganglion of Th. 3. The upper part of the lower third of it is about at the level of the dorsal root ganglion of Th. 7. And the abdominal esophagus is about at the level of the dorsal root ganglion of Th. 10 or Th. 11. Accordingly, degenerated nerve fibers following section of the dorsal root at points distal to their ganglia from Th. 3 to Th. 7 are found in the esophagus at each level of the dorsal root ganglia of Th. 3, Th. 7 and Th. 10 or Th. 11. The numbers of MARCHI'S granules were greatest at the level of the dorsal root ganglion of Th. 7.

I think these facts show that the visceral afferent fibers which have their cell bodies in a given dorsal root ganglion, are fairly widely distributed in the esophagus, mainly downward from the level of its ganglion. The state of their distribution seems to be similar to that of the visceral efferent fibers. LANGLEY stated that the fibers which enter the sympathetic trunk through a given white ramus may be distributed in from five to ten successive ganglia. GASKELL also pointed out similar facts in the dog.

MARCHI'S granules in the submucous tissue of the upper part of the lower third

of the thoracic esophagus are smaller in number than the granules in the same part following unilateral vagotomy. NOSE investigated physiologically the distributive density of afferent fibers of the vagus and the sympathetic nerve in the rabbit's abdominal esophagus, and said that the distribution of the vagus was more dense than that of the nerves from the sympathetic trunk from Th. 1 to Th. 10.

After phrenic exeresis, I did not find degenerated myelinated fibers in the portion of the esophagus in the diaphragm.

I have studied myelinated afferent fibers and have not referred to the unmyelinated fibers. The existence of the unmyelinated afferent fibers has not yet been determined.

### CONCLUSION

The secondary degeneration in the esophageal tissue following section of nerve trunks has been investigated mainly by MARCHI'S method and secondly by BIELSCHOWSKY-SETO'S method.

The following has been concluded.

(1) The esophagus is innervated afferently jointly by the vagus and the nerves which have their cell bodies in the dorsal root ganglia.

(2) In the esophagus, the distribution of the afferent fibers of the vagus is more dense than that of the afferent fibers which have their cell bodies in the dorsal root ganglia.

(3) The visceral afferent fibers which have their cell bodies in a given dorsal root ganglion, are fairly widely distributed in the esophagus, mainly downward from the level of its ganglion.

(4) Myelinated nerve fibers which enter the esophagus through the phrenic nerve are not found.

I am much indebted to Prof. Dr. KOH HIRAZAWA, Kyoto University Medical School for kindness to observe my stained preparations and also to Assistant Prof. Dr. CHUJI KIMURA of our clinic for his constant help throughout my study.

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## 和文抄録

## 犬食道の求心性二重神経支配に関する組織学的研究

京都大学医学部外科学教室第2講座 (青柳安誠教授 指導)

田 中 信 義

MARCHI 氏法を主とし、BIELSCHOWSKY - 瀬戸氏法を従として、迷走神経又は脊髄後根の切断による末梢神経の二次変性を犬食道組織内で検索して次の結果を得た。

- (1) 食道は迷走神経及び脊髄後根神経節内に神経細胞を有する神経により二重求心性神経支配を受けている。
- (2) 食道では迷走神経の求心性線維の分布濃度は脊髄後根神経節内に神経細胞を有する神経の求心性線維の

分布濃度より大である。

- (3) 一つの後根神経節内に神経細胞を有する内臓求心性線維は食道内では、その神経節の高さより主に下方にかなり広い範囲に分布している。
- (4) 横隔膜神経を経て食道に入る有髄神経線維は認められない。

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## 副腎皮質ホルモンの局所作用に対する年齢の影響

Effect of Age on Local Action of Adrenocortical Hormones

Pro. So. Ex. Bio. Med. V. 82 (No 2) 1953年

Burton L. Baker and Marjorie A. Schairer.

著者等は先に副腎皮質エキスを及び2, 3の副腎ステロイドを白鼠の皮膚に長期塗布していると手の成長が抑制され、幼弱白鼠の方が成熟白鼠よりも強く抑制されると云う実験を行つてゐる。今回の報告では其の実験方法を変更しているために反対の結果を出している。前回は幼弱白鼠も成熟白鼠も共に同一面積に同単位量の副腎皮質エキスを塗布したが、今回は両白鼠間の体表面積の相違を考慮に入れて皮膚の単位面積に同単位量の副腎皮質エキスを塗布するという方法を試みている。そして塗布する部は右頸背面で右耳と肩胛骨の中央と背中中央と体側中心線に囲まれた部分を用いている。即ち今回は塗布面積が大略々体面積に比例し、薬用量も又それに比例することになり幼弱白鼠では前実験よりも少量を用いた事になつてゐる。結果副腎皮質エキスの局所生長抑制作用は幼弱白鼠よりも成熟白鼠の方に強い事を見出している。

そして脳下垂体摘出白鼠において骨の生長に対して脳下垂体性生長ホルモンと副腎皮質エキスの拮抗作用をもつてゐることと、幼弱白鼠では末端組織中にこの生長ホルモンが成熟白鼠よりも多くあると考えられる事よりこの結果を説明している。(鈴江興二抄訳)